



TITLE Alternative uses for co-products: Harnessing the potential of valuable compounds from meat processing chains

AUTHORS Anne Maria Mullen, Carlos Álvarez, Dimitrios I. Zeugolis, Maeve Henchion, Eileen O'Neill, Liana Drummond

This article is provided by the author(s) and Teagasc T-Stór in accordance with publisher policies.

Please cite the published version.

The correct citation is available in the T-Stór record for this article.

NOTICE: This is the author's version of a work that was accepted for publication in *Meat Science*. Changes resulting from the publishing process, such as peer review, editing, corrections, structural formatting, and other quality control mechanisms may not be reflected in this document. Changes may have been made to this work since it was submitted for publication. A definitive version was subsequently published in *Meat Science*, 2017, Available online 3 May 2017, <https://doi.org/10.1016/j.meatsci.2017.04.243>

This item is made available to you under the Creative Commons Attribution-Non commercial-No Derivatives 3.0 License.



Alternative uses for co-products: Harnessing the potential of valuable compounds from meat processing chains

Anne Maria Mullen^{1*}, Carlos Álvarez¹, Dimitrios I Zeugolis^{2,3}, Maeve Henchion⁴, E. O'Neill⁵ & Liana Drummond¹

1.- Teagasc Food Research Centre,. Dept of Food Quality and Sensory Science. Ashtown, Dublin 15, Ireland

2.- Regenerative, Modular & Developmental Engineering Laboratory (REMODEL), Biomedical Sciences Building, National University of Ireland Galway (NUI Galway), Galway, Ireland

3. - Science Foundation Ireland (SFI) Centre for Research in Medical Devices (CÚRAM), Biomedical Sciences Building, National University of Ireland Galway (NUI Galway), Galway, Ireland.

4.- Teagasc Food Research Centre. Dept Agrifood Business and Spatial Analysis. Ashtown, Dublin 15, Ireland.

5.- University College Cork, Department of Food & Nutritional Sciences, Cork, Dublin

*Corresponding author: Anne.mullen@teagasc.ie. Tel: + 00 35318059521

Teagasc Food Research Centre, Ashtown, Dublin 15, Ireland

Abstract

Opportunities for exploiting the inherent value of protein-rich meat processing co-products, in the context of increased global demand for protein and for sustainable processing systems, are discussed. While direct consumption maybe the most profitable route for some, this approach is influenced greatly by local and cultural traditions. A more profitable and sustainable approach may be found in recognizing this readily available and under-utilized resource can provide high value components, such as proteins, with targeted high value functionality of relevance to a variety of sectors. Applications in food & beverages, petfood

biomedical and nutrition arenas are discussed. Utilization of the raw material in its entirety is a necessary underlying principle in this approach to help maintain minimum waste generation. Understanding consumer attitudes to these products, in particular when used in food or beverage systems, is critical in optimizing commercialization strategies.

Highlights

- Opportunities for extracting additional value from meat processing chain
- Meat co-products excellent source of proteins, vitamins, minerals and bio-actives
- High added-value uses can be developed to revalorize co-products

Keywords

Meat co-products, bioactive, protein, consumers, legislation, offal, valorisation, by-products

1. Introduction

Global pressures for increased supply of foods with high protein content, coupled with the ever increasing demand for more efficient processing and increased financial rewards, provides the imperative for the food industry to examine all processing streams with a view to adding or recovering value. Based on its amino acid profile, animal protein can provide a complete protein with a high biological value. In addition, meat, and in particular red meat, provides a rich source of macro and micro-nutrients. Demand for this high quality product is expected to continue to expand (FAO, 2009), which will lead to the increased production of the lower value ‘non meat’ products such as offal, blood, etc., which have been estimated to account for approximately 54-56% of the bovine and 48% of the porcine live animal weight (Marti, Johnson, & Mathews Jr, 2012; Vernooij, 2012). In this review, such non-meat products are referred to as co-products, similar to the view adopted in Australia. Classification as animal by-products (ABPs) is not always relevant as this classification under European legislation would restrict the use of these parts in the human food chain. While many countries have secured markets for them, in general these tend to be low value markets, often necessitating substantial transport costs. Even more, some of the products will carry a neutral (no disposal cost) or negative (costs money to dispose of) value. Making better use of the many co-products arising from meat processing, is not only important from a sustainability perspective, but also offers opportunities to the industry to increase their value.

As discussed by many authors (Lynch, Mullen, O'Neill, & Álvarez, 2017; Mullen, Álvarez, Pojić, Hadnadev, & Papageorgiou, 2015; Toldrá, Mora, & Reig, 2016) many of the materials are rich sources of valuable components such as protein, lipids, biomolecules etc. While some products command a reasonable value, and provide a nutritious food (Jayathilakan, Sultana, Radhakrishna, & Bawa, 2012) when consumed directly, for others it may be more valuable to extract separate components for discreet downstream applications. A five-stage universal recovery process was suggested by Galanakis (2012) to describe this process of transforming raw material into a final higher value product. While some of these steps can be eliminated or merged this gives a good overview of the process. Both traditional and emerging technologies of relevance are discussed in a later section. Optimal exploitation brings many challenges such as product hygiene and stabilisation, economies of scale, product purity, consumer attitudes and the requirement for efficient processing, which minimises the impact on the environment. These challenges can be addressed both through good industry practice and an applied research approach. Important principles to be guided by include that of total exploitation, avoiding the generation of additional waste (Waldron, 2007), and the cascading principle within the bioeconomy discourse which is about ensuring the highest value is extracted first with lower value options only considered once the higher ones have been exhausted.

Encouraging optimal utilisation of raw materials, early in the production chain, can help ameliorate food waste losses before the product becomes more dispersed along the food chain. The search for alternative protein sources has advanced in recent years (Van Huis et al., 2013), and provides a relevant approach to meeting global protein requirements. However, it is imperative to recognise that we have a readily available and under-utilised resource ripe for exploitation in co-products arising from meat processing. As there is a link between meat consumption and demand for protein-rich feed for livestock (Pimentel & Pimentel, 2003; Trostle, 2008), it is imperative that all meat processing related proteins are optimally exploited. This review covers applications relating to use in food and beverage systems, petfood, nutrition focused and bioactive & biomedical applications. Market and legislative information is provided in addition to an overview of relevant technologies and consumer attitudes.

2. Applications

2.1 Food ingredients

Among other attributes proteins can impart techno-functional properties within a food or beverage system. For example they may influence water holding capacity, viscosity, gelation, emulsification and foaming. The ability of proteins to function in this manner is determined by intrinsic e.g. amino acid profile, molecular structure and surface hydrophobicity, and by extrinsic factors such as pH temperature, and ionic strength. Collagen, a fibrous, structural protein, is one of the most abundant proteins. It is probably the animal protein most regularly employed in food production. Following extraction and hydrolysis it can be transformed into gelatine whose strong gelling properties are relevant to a variety of products such as soups, gravies, desserts or dairy products (Hettiarachchy, Sato, Marshall, & Kannan, 2012). Gelatine is mainly extracted from skin and bones. However, other offal such as lung, tongue, trachea, large blood vessels, or tendons are, also, sources of collagen. Collagen has very good film forming capacity which is exploited to generate casings and novel packaging (Gómez-Guillén, Giménez, López-Caballero, & Montero, 2011). Other more specific applications of collagen are described below in section 2.4, and have also been reviewed by various authors (Ferraro, Anton, & Santé-Lhoutellier, 2016; Zeugolis, Paul, & Attenburrow, 2009). A variety of commercial collagen based products are available for use in food systems. Examples of companies supplying them include Rousselot, Collapro and Devro, who provide both clean label binders, gelling and textural agents or collagen based sausage casing. Blood plasma comes second in line in meat co-product derived protein most employed as a food ingredient. Following hygienic collection, centrifugation leads to the recovery of plasma and cellular fractions (Lynch et al., 2017). Plasma can be used as it is, or dehydrated for storage as a dry powder (Pares, Toldra, Saguer, & Carretero, 2014). Blood plasma is a versatile product, which presents good emulsifying, gelling, foaming and solubility properties. Applications of plasma in food industry see it used as a binder in meat products, egg replacers in bakery, protein-rich pasta, fat replacers or even polyphosphate substitute (Hsieh & Ofori, 2011; Hurtado et al., 2011). Some examples of plasma proteins currently in the market are: Fibrimex® (purified fibrinogen used as binder), Immunolin® (concentrated immunoglobulins, which improve the immune system)(Abu-Akkada & Awad, 2015), Myored (natural red colorant), Vepro 95 HV (purified globin used as emulsifier) or Plasma Powder FG (plasma with increased content of fibrinogen used as cold binder) (Ofori & Hsieh, 2012). Apart from those two sources, the use of offal as sources of proteinaceous food ingredients has not received a lot of research funding. In the last decade research has been published describing the extraction and characterisation of functional proteins from various offal such as: liver (Steen et al., 2016; Zou et al., 2017), lung (Selmane, Christophe, & Gholamreza,

2008), heart (Dewitt, Gomez, & James, 2002), viscera (Bhaskar, Modi, Govindaraju, Radha, & Lalitha, 2007) or bones. (Linder, Fanni, Parmentier, Sergeant, & Phan-Tan-Luu, 1995). Very few patents, using offal as an ingredient, have been filed in the last 20 years; for instance a patent filed in 2001 relates the use of offal as an ingredient for cheaper sausages or burger patties (Van, 2001). But, in spite of the advances generated in food processing, most of the patents found in the literature are from the 1960's and 1970's.

At a very basic, low value level, protein extracts from offal can be used as extender in processed meat products (Heinz & Hautzinger, 2007). Recently, the use of pork tongue and pork head meat as sausage ingredients has been reported, with no significant differences in some quality characteristics compared to controls (Choi, Hwang, et al., 2016). Similar results were observed when the final products were hamburger patties (Choi, Jeon, et al., 2016). However, considering the functional properties of many of these proteins, there is a strong argument to be made for capitalising on this high-value potential to more fully support a sustainable use of natural resources. It is important to also remember that, as well as providing proteins with nutritive value or good techno-functional characteristics, these co-products are well established as being rich in minerals and vitamins (Mullen & Álvarez, 2016).

2.2 Nutritional quality of co-products

Nutritional quality of proteins has traditionally been defined based on the amino acid profile, and the ability to provide specific patterns of amino acids for protein synthesis as measured by animal growth or nitrogen balance in humans (Millward, Layman, Tomé, & Schaafsma, 2008). However, a more current definition describes this term as the ability of the protein to achieve specific metabolic activities. Currently, the objective of protein quality evaluation is determining the ability of a protein to reach maintenance needs, along with special needs as growth, pregnancy or lactation (WHO, 2007). The up-to-date method approved for protein quality determination is the protein digestibility-corrected amino acid score (PDCAAS). In this method the single most limiting amino acid is identified, when compared to an appropriate reference pattern, which defines the amino acid score. According to FAO, Lysine, Methionine and Tryptophan are the most limiting amino acids in poor protein sources. Table 1 shows the amount of these three amino acids, the total protein content and the percentage of essential amino acids in a variety of offal from pork, beef and lamb. It can be seen that offal, generally, is a good source of essential (namely Ile, Leu, Lys, Met, Phe, Thr, Trp, Vali) and limiting amino acids. However, offal rich in connective tissue as ears,

feet, or lips, present a lowest protein quality, since connective tissue is mainly composed by glycine, proline and alanine, which are not essential amino acids (Mullen & Álvarez, 2016). This lack of essential amino acids can be overcome by tailor blending ingredients to achieve a balanced amino acid profile in the final product.

Besides proteins, meat co-products can be a good source of minerals (Ca, Fe, Mg, Cu or Se) or vitamins (niacin, Vit. B12, folate or Vit. C). Heme iron, mainly provided by meat and fish, is more absorbable than non-heme iron (<15%); and also, although the mechanism is not well defined, heme iron is more absorbed in the presence of meat, what is known as the “meat factor” (López & Martos, 2004). Thus, it is worthy to explore if protein derived from offal can also increase the bioavailability of iron, since they are a good iron source. Finally, the offal fat content can vary from less than 1% in blood, up to 17% in tongue; with high contents of saturated fatty acids. Among fats, it is important to draw attention to cholesterol when referring to co-products, as some have high levels of this compound, as for example kidneys, liver or brain (Mullen & Álvarez, 2016).

It is clear that meat co-products are rich sources of macro and micro-nutrients. Both these raw materials and various extracts generated from them can be incorporated in products for targeted end-users such as the elderly, sports-active or dairy intolerant consumers (Montero Castillo, Ligardo, Alejandro, González, & Cristina, 2015).

2.3 Bioactive compounds

Bioactive compounds cover both peptides and other biomolecules which exert a physiological benefit on consumption. Bioactive peptides or biopeptides are defined as short sequences of 2 to 30 amino acids that exert this effect. These sequences are encrypted in the parent protein and must be released from the parent protein to have an effect (Di Bernardini, Harnedy, et al., 2011; Ryan, Ross, Bolton, Fitzgerald, & Stanton, 2011). In order to release these encrypted amino acid sequences, three different techniques are employed: enzymatic hydrolysis by means of commercial proteases (Sarmadi & Ismail, 2010); proteolytic microorganism; or after fermentation or aging processes (Escudero et al., 2013). After any of these processes, hundreds of peptides are generated, but only a few of them show biological activity. For this reason, a further purification step must be carried out. Techniques such as membrane filtration, chromatography or differential precipitation are commonly employed (Power, Fernández, Norris, Riera, & FitzGerald, 2014). A different approach is to firstly isolate the proteins, and then, carry out the hydrolysis step, which makes the further isolation process easier. In order to optimise the screening process for bioactive peptide searching, *in*

silico tools can be employed (Fu et al., 2016): these are useful to find the right protein/enzyme combination. As a complementary tool, the peptides of interest can be synthesized at lab scale, in such way further studies of toxicology, functionality, absorption, bioavailability or dose response can be performed (Korhonen & Pihlanto, 2006).

As discussed, the main component of meat co-products, on a dry basis, is protein; thus, these by-products are a potential source of bioactive peptides which can be extracted, purified and commercialised as a very high-added value product (Lemes et al., 2016). An ideal scenario is one where proteins with key techno-functional characteristics are first extracted from the raw material and the residual proteinaceous material subjected to biopeptide generation. An example of this is seen on the ReValueProtein project (Álvarez, Lynch, Drummond, & Mullen, 2016) where techno-functional proteins are extracted from lung and the non-solubilised material used to generate bioactive peptides. Compared to other source materials relatively few studies have looked at generating bioactive peptides from meat co-products.

Examples of bioactive peptides obtained from blood and meat are published or reviewed by (Bah, Bekhit, Carne, & McConnell, 2013; Di Bernardini, Harnedy, et al., 2011; Toldra, Aristoy, Mora, & Reig, 2012). However, a more limited number of studies can be found using offal as a source (Di Bernardini, Rai, et al., 2011; Lee et al., 2010; O'Sullivan, Lafarga, Hayes, & O'Brien, 2016). The types of activities reported have included antioxidant, antimicrobial, antihypertensive, anti-diabetic, anti-hypercholesterolemic or mineral binding (Sharma, Singh, & Rana, 2011). A number of important points need to be addressed to fully capitalise on the value of these peptides. Ensuring that the peptides reach the target organ, are non-toxic and both bioavailable and bioactive in the body is critical. The scale of processing and the economic feasibility of same are also key considerations: which can be somewhat mitigated by readily available, low price raw materials combined with novel hydrolysis and purification steps (Agyei & Danquah, 2011).

Other molecules of interest, as health promoting compounds include creatine, carnosine, carnitine, anserine and taurine. These naturally occurring low molecular weight molecules can be found mainly in skeletal muscles and have many positive effects as: antioxidant, lowering cholesterol, improved calcium absorption, preventing muscle myopathy, or preventing heart disease (Williams, 2007) hence exudate and cook-out may be good sources. It has also been reported that heart or liver can be a good source of these compounds (Hoffmann, Waszkiewicz-Robak, & Świdorski, 2010). Though not the subject of this review, many other active compounds can be found in meat and may also be present in co-products, such as: conjugated linoleic acid (CLA), chondroitin sulphate, Coenzyme Q10, spermidine,

choline, lipoic acid, or glutathione (Arihara & Ohata, 2008). The main biological activities are summarized in Table 2.

2.4 Biomedical applications

Natural biomaterials such as collagen present many opportunities outside of food sector and in particular can serve as raw materials in biomedical applications. The term ‘collagen’ encompasses a family of glycoproteins that are characterised by a repeating [Glycine-X-Y]_n amino acid sequence (X is usually proline and Y is frequently hydroxyproline). To-date, twenty-nine genetically distinct collagen types have been described that can be classified in four broad categories [fibrous (e.g. types I, II, III), non-fibrous (e.g. types IV, VII, XXVIII), filamentous (e.g. types VI, VIII, X) and fibril associated with interrupted triple helices (e.g. types IX, XII, XIV) collagens], based on their primary structure, molecular weight, charge profile along the helix, length of the triple helix, size and shape of the terminal globular domains. In vertebrates, collagen is the major constituent of connective tissues, comprising almost 25 % of total body proteins, 75 % of the dry weight of skin, 80 % of the organic matter in bones and 90 % of tendon and corneal tissues. The abundance of this extracellular matrix protein, in addition to its inherent bioactivity, molecular recognition signals, controllable mechanical and degradation properties and its ability to be reconstructed in various three-dimensional architectures (e.g. fibres, sponges, films, spheres, hydrogels), make it an ideal raw material for biomaterials, tissue engineering and drug / gene / cell delivery applications (Abbah et al., 2015; Kielty & Grant, 2002; Soroushanova et al., Submitted; Thomas et al., 2016).

In biomedicine, porcine and bovine skin and tendon tissues are the main source of collagen type I, whilst collagen type II is extracted from porcine and bovine articular cartilage. Fish industry by-products (e.g. skin, scales and bones) have also been used to extract collagen for the fabrication of biomaterials, but to a smaller extent. Independently of the source (e.g. mammalian or fish), purified collagens are extracted using primarily dilute acids with or without enzymes. Dilute acids disassociate mild intermolecular aldimine cross-links, whilst proteolytic enzymes (e.g. pepsin) are effective even against stable ketoimine bonds, not only increasing the yield for up to 10 times, but also lowering immune response in patients, through the removal of the antigenic sequence P-determinant, located at the telo-peptide regions (Bruckner & Prockop, 1981; Lynn, Yannas, & Bonfield, 2004; D. I. Zeugolis, Paul, & Attenburrow, 2008). While collagen and collagen-like molecules can be produced using various cell types, recombinant systems and peptide synthesis, these technologies are still

very expensive, of low yield, and produce unstable collagens. Compared to animal extracted collagen they have increased susceptibility to proteolytic degradation, restricting their use in biomedicine (Browne, Zeugolis, & Pandit, 2013; Fichard, Tillet, Delacoux, Garrone, & Ruggiero, 1997; Frischholz et al., 1998; Myllyharju et al., 1997; Vuorela, Myllyharju, Nissi, Pihlajaniemi, & Kivirikko, 1997). All these factors support continued expansion of the use of collagen in the biomedicine, food and cosmetic industry.

2.5 Pet food and animal feed

As nutritious sources of protein, fat and micronutrients many products are highly suitable for use in companion animal diets (Cramer et al., 2007; Rivera, 1998). As rich sources of protein and as a result of regular use in pet food formulations they have been considered a major contributor to the growth of the pet food industry (Corbin, 1992). In canine diets they provide good sources of digestible nutrients in particular proteins and fat (Murray et al 1997). Palatability is an important consideration in pet foods formulation. Co-products can also serve as palatability enhancers (Boskot, 2009) which will have a direct impact on product acceptability. The pet food market has seen strong growth with pet humanisation and premiumisation being considered key drivers. As well as being incorporated into petfoods these products are also sold in pet stores as treat type products e.g. bone, hides, ears etc. Animal feeds play an important role in the global food industry as they influence greatly the ability to produce animals in an economical way. In particular, following rendering, co-products arising from meat processing form an important constituent in animal feeds (Jayathilakan, Sultana, Radhakrishna, & Bawa, 2012).

3. Market overview

The categorization and utilization of a secondary stream as a co-product is dependent on several factors such as local and export market demands, existing profitable applications and a favourable and feasible commercialization route. Broadly speaking, depending on local cultural habits, the market for 5th quarter products such as offal and blood is shared between exports, pet food, animal feed and local direct consumption.

Clear opportunities for exploring a variety of meat co-products have been identified over the years in scientific research studies and market trend reports. However, an effective market analysis of this area needs to take account not only of the latest R&D and scientific advances in the recovery and reutilization of compounds from co-product materials, but also of industry activities currently in place, as well as existing market and commercial scenario of

the original products, since re-direction of a stream may have impacts on established supply chains and ancillary commercialization routes. Several markets with existing or future potential based on co-products are briefly presented below.

The protein market is generally divided by applications: food and beverage, sports nutrition, animal feed, cosmetics and personal care, pharmaceuticals, etc. For the sports nutrition category in particular, protein stands as the main driving force (Schmidt, 2014). In Ireland and other markets, protein powders are still the most popular type of sports nutrition product, but other more convenient formats such as protein ready-to-drink products and protein bars are quickly becoming more popular (Hickey, 2014). As reported by Glanbia “the global performance nutrition market, at retail selling price, is approximately \$10.1 billion with the USA accounting for 63%, and other international markets accounting for 37%” (Glanbia.com, 2017). Although whey proteins still dominate this market, due to diversification of products and applications, blends incorporating other sources of proteins are increasingly gaining ground.

The market size for functional foods may be difficult to measure, as the exact definition of a functional food can vary between regions, countries or even companies. Nonetheless, some analysts value it at up to USD 190 Bn worldwide, showing a growing of 6–25% per year, and with the same perspective for the next 5 years (Calvo, Martorell, Genovés, & Gosálbez, 2016).

The global collagen market is forecast to expand at a 9.4 % compound annual growth rate from 2015 to 2023 and the market is projected to rise to US\$ 9.37 billion by 2023 ("Collagen market - Global industry analysis, size, share, growth, trends, and forecast 2015 – 2023," Transparent Market Research Report 2016). Currently, around 60% is due to gelatine production (Global Market Insights report, 2015). Other markets which go beyond food and beverage include the use of collagen for biomedical applications and the manufacture of advanced biomaterials, for tissue regeneration for example, an area reported to be expanding rapidly (Global Market Insights report, 2015). Collagen has also attracted much interest as a source of functional peptides, with applications in the health, cosmetics and personal care areas.

The challenges in obtaining approval for peptides as ingredients with beneficial health properties has undoubtedly influenced the growth of this market, but for products which receive approval there are clear rewards. In 2012 while approved peptide drugs were estimated to command only about 2% of the global drug market the value represented approximately USD 20 billion (Sun, 2013).

Key markets for animal blood include the food industry, animal feed and pet food applications. It is estimated that the food industry utilizes about 30% of blood produced from slaughter in different forms, for example plasma used as an emulsifier or whole blood used in traditional products (Bah et al., 2013). The value of the US dried animal blood plasma market for animal feed in 2010 was estimated in USD 29 million per annum (Wilson, 2011). The possibility of obtaining bioactive peptides from blood could lead to higher value products being produced, giving access to the expanding functional and nutraceutical applications market. Further efforts for the extraction and valorisation of protein from animal blood may also increase the use of blood as a source of techno-functional compounds for food and beverage applications.

While the export market for some 5th quarter products offer the greatest opportunity to increase margins from edible offal, consumer perceptions in relation to food scares or disease outbreaks may have an unjustifiable impact of product values. Additionally, an understanding of specific market requirements is necessary, to satisfy demands in terms of customs and expectations.

4. Consumer studies

Humans seek novelty and variety in their diet for nutrition and health reasons and to satisfy pleasure seeking motives (Al-Shawaf, Lewis, Alley & Buss, 2015). However they are also wary of foods with which they are unfamiliar, due to fears in relation to food safety and health. This led to the phrase “the omnivores dilemma” to reflect individual’s competing motives of neophobia and neophilia (Martins, Pelchat & Pliner, 1997). Reflecting differences in cultures, meat co-products are regarded as delicacies in some countries, however most consumers in developed countries are unfamiliar with meat co-products, such that many have never tasted offal and most have a very limited knowledge about how to prepare and cook it. Some may even regard them as unhealthy or waste (Frewer & Gremmen, 2007).

Three important factors may lead to the rejection of unfamiliar food products: negative sensory properties, harmful consequences and “ideational” factors (Rozin & Fallon, 1987). Henchion et al (2016) found such factors at play in consumer evaluations of a range of product concepts that incorporated ingredients derived from offal. On the negative side, consumers were concerned with the taste and texture of such products, did not see significant benefits, were disgusted with the idea that the ingredients originated in live animals and even questioned if such products were edible. However this research also found that processing could have a positive influence on acceptance through its influence on the physical form of

the ingredient. Changing the physical form of the ingredient resulted in “de-animalising” the ingredient and for some consumers shifted the focus away from emotive, ideational factors, and allowing them to consider the product’s characteristics and benefits. The impact of processing is however complex with increased processing resulting in negative perceptions regarding healthiness and naturalness in some contexts.

Integrating unfamiliar foods into existing culinary and dietary practices was identified as supporting acceptance in the context of insect-based foods (Looy, Dunkel, & Wood, 2014). Research by Henchion et al (2016) reports similar findings with regards to meat co-products suggesting that this strategy could also work for meat co-products. They argue that meat co-products should be presented to consumers as ingredients rather than finished products to give them flexibility in terms of how they are integrated into consumers’ routines.

The role of experts, celebrity chefs and friends in providing evidence that consumption of unfamiliar products is safe and socially acceptable is highlighted in relation to meat co-products in some niche markets. Boutique butchers with traditional “fancy meats” and restaurants featuring nose-to-tail eating are becoming more prevalent in the UK for example, resulting in changes in the consumer landscape, with a claim by Datamonitor (2014) that “offal is officially in”, due to experimentation, consciousness of value and increased availability.

5. Legislation

To explore and commercialise the range of available co-products of animal origin it is necessary to understand relevant legislation, rules and regulations governing access to source materials and markets. Information and guidance on how material is to be collected, handled, stored, processed, etc. will determine not only the value of the final product, but crucially ensure compliance for downstream uses. It can be said that legislation was developed for two distinct groups: unusable and usable by-products (Leoci, 2014). For unusable by-products, associated regulations lay down handling and disposal rules to safeguard the environment and public health. On the other hand, for by-products posing no risk to health or the environment, and for which a viable commercial application exists, regulations may relate to materials’ collection, transport and further processing.

In the European Union, the term animal by-product (ABP), is reserved for products of animal origin that are not intended for human consumption and use or disposal of ABP is strictly controlled. Depending on the risk they pose, ABPs are divided into categories 1, 2 and 3, category 1 being the highest in risk. The classification of some 5th quarter products as edible

or inedible can be influenced by collection and handling conditions and on local or existing export markets. If intended for human consumption, 5th quarter products are covered under regulations for food of animal origin, and should not be classified as ABPs.

Since the bovine spongiform encephalopathy (BSE) crises, rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies (TSEs) set out in Regulation 999/2001, have irreversibly excluded parts of the carcass from the food chain. The regulation covers the production, placing on the market and, in some cases, the export of animals and animal products. For feed and pet food, for example, only category 3 material is allowed. Directive 2008/98/EC on waste includes a definition and the main conditions which must be met by a substance to be classified as a by-product. The regulation encourages the reuse, reutilization and recycling of non-hazardous by-products. Also relevant, Directive 2008/1/EC prohibits discharging of blood into digesters and sets down an obligation of using blood for other purposes.

Regulation (EC) No 1069/2009 includes health rules as regards animal by-products and derived products not intended for human consumption (uses other than for food). However, some products are not covered by this Regulation. These include cosmetic products (Directive 76/768/EEC); medical implants (Directive 90/385/EEC); medical devices (Directive 2003/32/EC); In vitro diagnostic medical devices (Directive 89/79/EC); veterinary medical products (Directive 2001/82/EC); and medicinal products (Directive 2001/83/EC).

For parts of the carcass intended for human consumption, the "Hygiene Package": Regulation (EC) 852/2004, Regulation (EC) 853/2004 and Regulation (EC) 854/2004), must apply, as well as the Regulation (EC) 1169/2011 for the provision of food information to consumers. From the perspective of food hygiene regulations, offal is defined as: "fresh meat other than that of the carcass, including viscera and blood" (EC) 854/2004). However, from an ingredient labelling perspective, Regulation (EC) 1169/2011 restricts the definition of "meat" to skeletal attached muscles. Any other parts of the animal must be declared separately and the meat species must be identified on the label (e.g.: "beef heart protein" or "bovine heart protein"), which can have an impact on consumer perception of these food ingredients.

Also relevant, assuming they are utilised in products for these target audiences, Regulation (EU) No 609/2013 on food intended for infants and young children, food for special medical purposes, and total diet replacement for weight control, lays down general rules on the composition and preparation of foods that are specially designed to meet the particular nutritional requirements of the persons for whom they are intended.

Regulation 1333/2008 (latest consolidated version published February 2016) covers food additives and lists substances that are not considered as additives, and hence not covered by this regulation, which includes blood plasma, protein hydrolysates and their salts. These are also not considered meat and must be declared separately on the label. Annex II to Regulation 1333/2008, amended by Regulation (EU) 601/2014, lays down a list of approved food additives and their conditions of use. Food additives are listed on the basis of the categories of food to which they may be added to, with Category 8 covering processed and unprocessed meat. Food enzymes are covered separately by Regulation (EC) 1332/2008.

In the United States, Title 9 of the Code of Federal Regulations (CFR) provides the rules governing animals, and animal products. It includes two groups of protein contributing ingredients: Group 1 includes ingredients of livestock or poultry origin from muscle tissue which are skeletal or found in the edible organs, with or without the accompanying and overlying fat, and the portions of bone, skin, sinew, nerve, and blood vessels which normally accompany the muscle tissue, not separated in the process of dressing, as well as meat by products; mechanically separated (species); and poultry products. Ingredients processed by hydrolysis, extraction, concentrating or drying are part of Group 2 Protein-Contributing Ingredients along with any other ingredient which contributes protein. An up-to-date list of substances considered as safe and suitable ingredients for the production of meat, poultry and egg products is published in a regularly revised directive from the USDA-FSIS, currently Revision 39, from January, 2017. The list includes beef protein as a food grade substance approved as generally recognized as safe (GRAS), for use in meat, poultry and egg products. Federal regulation (USDA-FSIS, 1999) lays down the rules and regulations for collection and processing of blood when intended for human consumption.

Meat protein ingredients are considered non-allergenic and are generally “minimally processed”. Additionally, their nomenclature is more easily recognised. Although this can make them more consumer-friendly, it may also negatively affect consumer perception. Table 3 summarizes the current legislative scenario in Europe, regarding the use of meat co-products.

6. Relevant technologies

Traditional methods such as chemical precipitation, ultrafiltration, extrusion, lyophilisation, isoelectric solubilization-precipitation or solvent extraction are widely employed in order to extract and concentrate proteins and peptides from waste materials (Galanakis, 2015). Depending on the raw material, and the final application of the extract, one of these methods, or a combination of them, has to be selected. However, some of the methods above

mentioned, may have a negative impact on the activity of the desired compounds, since heat treatment (for instance evaporation, pasteurization or extrusion) or denaturing chemicals (ethanol, strong acids or alkalis) might irreversibly affect the structure and chemical properties of the target compounds. In the particular case of proteins and peptides, negative processing effects are (Korhonen, Pihlanto-Leppäla, Rantamäki, & Tupasela, 1998):

- i) Heat treatment: destruction of heat sensitive peptides, cross linkages, protein denaturation
- ii) pH: racemization, destruction of amino acids, risk of oxidation
- iii) Membrane filtration: change in amino acid composition
- iv) Storage: destruction of lysine and oxidation.

These changes can lead to a loss of protein functionality, nutritive value or bioactivity; reducing the final value of the product and narrowing their field of application. In order to minimise or prevent such modifications, novel technologies are being developed. Such technologies usually overcome the negative impacts of thermal treatments and additionally, minimise significantly the amount, and hence the effect, of solvents and reagents employed for protein/peptide extraction.

Most promising and emerging technologies for protein extraction are those based on ultrasounds (Kadam, Tiwari, Álvarez, & O'Donnell, 2015); pulsed electric fields (PEF) (Soliva-Fortuny, Balasa, Knorr, & Martín-Belloso, 2009); high hydrostatic pressures (HHP) (Li, Zhu, Zhou, & Peng, 2012); sub-critical water hydrolysis (SWH) (Marcet, Alvarez, Paredes, & Diaz, 2016); or laser ablation (Boutinguiza et al., 2007). Among these techniques, ultrasounds and PEF have been reported to produce negligible changes in protein composition and structure (Chandrapala, Zisu, Palmer, Kentish, & Ashokkumar, 2011; Garde-Cerdán, Arias-Gil, Marsellés-Fontanet, Ancín-Azpilicueta, & Martín-Belloso, 2007). On the other hand, HHP and SWH, can produce drastic changes in protein structure as non-reversible unfolding or hydrolysis (Smeller, 2002). Other technologies which hold potential for these products include aqueous two phase extraction, foam mat drying, and others reported by Galanakis (2012). Table 4 presents how, through the application of currently available technologies, is possible to obtain high added-value products from the various co-products generated by the meat industry.

There is little evidence of the application of many of these emerging technologies for extracting value from meat co-products. More effort has been seen in optimizing the use of these emerging technologies on vegetables, seaweeds and dairy products. There clearly is a pressing need to examine the relevance of transferring these developments into the meat

sector in terms for example of both recovery yields and impact on functionality of final product.

7. Conclusions

Opportunities clearly exist for extracting additional value from meat processing chains. As well as ensuring optimal use of such a protein rich material this approach also supports sustainable practices across the sector sector and will be increasingly necessary in a world of growing demand and constrained resources. Science and technology driven approaches must be coupled with market, legislative and consumer knowledge for successful commercialisation of the downstream products.

Acknowledgements:

This work forms part of the ReValueProtein Research Project (Grant Award No. 11/F/043) which is supported by the Irish Department of Agriculture, Food and the Marine (DAFM) and the Food Institutional Research Measure (FIRM) both funded by the Irish Government under the National Development Plan 2007-2013.

8. Bibliography

- Abbah, S., Delgado, L., Azeem, A., Fuller, K., Shologu, N., Keeney, M., . . . Zeugolis, D. (2015). Harnessing hierarchical nano- and micro-fabrication technologies for musculoskeletal tissue engineering. *Adv Healthc Mater*, 4(16), 2488-2499.
- Abu-Akkada, S. S., & Awad, A. M. (2015). Protective effects of probiotics and prebiotics on *Eimeria tenella*-infected broiler chickens. *Pak. Vet. J*, 35, 446-450.
- Agyei, D., & Danquah, M. K. (2011). Industrial-scale manufacturing of pharmaceutical-grade bioactive peptides. *Biotechnology Advances*, 29(3), 272-277.
- Al-Shawaf, L., Lewis, D. M. G., Alley, T. R. & Buss, D. M. (2015). Mating strategy, disgust, and food neophobia, *Appetite*, 85, 30-35.
- Álvarez, C., Lynch, S., Drummond, L., & Mullen, A. M. (2016). A multidisciplinary approach for the recovery and revalorisation of proteins from bovine and porcine co-products. [Oral communication]. *Congreso Iberoamericano de Biotecnología*, 1(1), 195.
- Arihara, K., & Ohata, M. (2008). Bioactive compounds in meat *Meat biotechnology* (pp. 231-249): Springer.
- Bah, C. S. F., Bekhit, A. E.-D. A., Carne, A., & McConnell, M. A. (2013). Slaughterhouse Blood: An Emerging Source of Bioactive Compounds. *Comprehensive Reviews in Food Science and Food Safety*, 12(3), 314-331.
- Bhaskar, N., Modi, V., Govindaraju, K., Radha, C., & Lalitha, R. G. (2007). Utilization of meat industry by products: protein hydrolysate from sheep visceral mass. *Bioresource technology*, 98(2), 388-394.
- Boskot, S. (2009) Production of pet food: inclusion of palatability enhancers in dry extruded dog food. Lambert Academic Publishing AG & Co KG, Germany.

- Boutinguiza, M., Lusquiños, F., Comesaña, R., Riveiro, A., Quintero, F., & Pou, J. (2007). Production of microscale particles from fish bone by gas flow assisted laser ablation. *Applied Surface Science*, 254(4), 1264-1267.
- Browne, S., Zeugolis, D. I., & Pandit, A. (2013). Collagen: Finding a solution for the source. *Tissue Eng A*, 19(13-14), 1491-1494.
- Bruckner, P., & Prockop, D. J. (1981). Proteolytic-enzymes as probes for the triple-helical conformation of procollagen. *Anal Biochem*, 110(2), 360-368.
- Calvo, D. R., Martorell, P., Genovés, S., & Gosálbez, L. (2016). Development of novel functional ingredients: Need for testing systems and solutions with *Caenorhabditis elegans*. *Trends in Food Science & Technology*, 54, 197-203.
- Chandrapala, J., Zisu, B., Palmer, M., Kentish, S., & Ashokkumar, M. (2011). Effects of ultrasound on the thermal and structural characteristics of proteins in reconstituted whey protein concentrate. *Ultrasonics Sonochemistry*, 18(5), 951-957.
- Choi, Y.-S., Hwang, K.-E., Kim, H.-W., Song, D.-H., Jeon, K.-H., Park, J.-D., . . . Kim, C.-J. (2016). Replacement of Pork Meat with Pork Head Meat for Frankfurters. *Korean Journal for Food Science of Animal Resources*, 36(4), 445.
- Choi, Y.-S., Jeon, K.-H., Ku, S.-K., Sung, J.-M., Choi, H.-W., Seo, D.-H., . . . Kim, Y.-B. (2016). Quality characteristics of replacing pork hind leg with pork head meat for hamburger patties. *Korean journal of food and cookery science*, 32(1), 58-64.
- Collagen market - Global industry analysis, size, share, growth, trends, and forecast 2015 – 2023. (2016): Transparency Market Research.
- Corbin, J.E. (1992) Inedible meat, poultry and fish by-products in pet foods. In Pearson, A.M. and Dutson, T.R. (eds) Inedible meat by-products. *Advances in meat research*, Vol 8. Pp 329-347, Elsevier Applied Science, London.
- Cramer, K. R., Greenwood, M. W., Moritz, J. S., Beyer, R. S. & Parsons, C. M. (2007) Protein quality of various raw and rendered by-product meals commonly incorporated into companion animal diets. *Journal of Animal Science*. 85:3285–3293.
- Datamonitor. (2014) Consumer and Innovation trends in Meat Fish and Poultry 2014. Available at: http://www.datamonitor.com/store/Product/consumer_and_innovation_trends_in_meat_fish_and_poultry_2014?productid=CM00198-064. Accessed February 2017
- Dewitt, C., Gomez, G., & James, J. (2002). Protein extraction from beef heart using acid solubilization. *Journal of food science*, 67(9), 3335-3341.
- Di Bernardini, R., Harnedy, P., Bolton, D., Kerry, J., O'Neill, E., Mullen, A. M., & Hayes, M. (2011). Antioxidant and antimicrobial peptidic hydrolysates from muscle protein sources and by-products. *Food Chemistry*, 124(4), 1296-1307.
- Di Bernardini, R., Rai, D. K., Bolton, D., Kerry, J., O'Neill, E., Mullen, A. M., . . . Hayes, M. (2011). Isolation, purification and characterization of antioxidant peptidic fractions from a bovine liver sarcoplasmic protein thermolysin hydrolyzate. *Peptides*, 32(2), 388-400.
- Escudero, E., Mora, L., Fraser, P. D., Aristoy, M.-C., Arihara, K., & Toldrá, F. (2013). Purification and identification of antihypertensive peptides in Spanish dry-cured ham. *Journal of proteomics*, 78, 499-507.
- EC Regulation 999/2001. Rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies. *Official Journal of the European Union*, 147, 1–69
- EC Regulation 853/2004. Specific hygiene rules for on the hygiene of foodstuffs. *Official Journal of the European Union*, 139, 1–55.
- EC Directive 2008/98/EC on waste and repealing certain Directives. *Official Journal of the European Union*, 312, 3-30
- EC Directive 2008/1/EC concerning integrated pollution prevention and control. *Official Journal of the European Union*, 24, 8-29
- EC Regulation 1333/2008. Food additives. *Official Journal of the European Union*, 354, 16- 33

EC Regulation 1069/2009. Health rules as regards animal by-products and derived products not intended for human consumption. Official Journal of the European Union, 300, 1–33

EC Regulation 1169/2011. Provision of food information to consumers. Official Journal of the European Union, 304, 18-62

EC Regulation 609/2013. Food intended for infants and young children, food for special medical purposes, and total diet replacement for weight control. Official Journal of the European Union, 181, 35-56.

Fallas, J. A., O'Leary, L. E., & Hartgerink, J. D. (2010). Synthetic collagen mimics: Self-assembly of homotrimers, heterotrimers and higher order structures. *Chem Soc Rev*, 39(9), 3510-3527.

FAO. How to Feed the World in 2050. (2009). Available from: http://www.fao.org/fileadmin/templates/wsfs/docs/expert_paper/How_to_Feed_the_World_in_2050.pdf. Accessed on February 2017.

Ferraro, V., Anton, M., & Santé-Lhoutellier, V. (2016). The “sisters” α -helices of collagen, elastin and keratin recovered from animal by-products: Functionality, bioactivity and trends of application. *Trends in Food Science & Technology*, 51, 65-75.

Frewer, L.J. & Gremmen, B. (2007). Consumer's interests in food processing waste management and co-product recovery. In: Waldron K (Ed) Handbook of waste management and co-product recovery in food processing, vol. 1. Cambridge, UK: Woodhead Publishing, 21-33.

Fichard, A., Tillet, E., Delacoux, F., Garrone, R., & Ruggiero, F. (1997). Human recombinant alpha1(V) collagen chain. Homotrimeric assembly and subsequent processing. *J Biol Chem*, 272(48), 30083-30087.

Frischholz, S., Beier, F., Girkontaite, I., Wagner, K., Pöschl, E., Turnay, J., . . . von der Mark, K. (1998). Characterization of human type X procollagen and its NC-1 domain expressed as recombinant proteins in HEK293 cells. *J Biol Chem*, 273(8), 4547-4555.

Fu, Y., Young, J. F., Løkke, M. M., Lametsch, R., Aluko, R. E., & Therkildsen, M. (2016). Revalorisation of bovine collagen as a potential precursor of angiotensin I-converting enzyme (ACE) inhibitory peptides based on in silico and in vitro protein digestions. *Journal of Functional Foods*, 24, 196-206.

Galanakis, C. M. (2012). Recovery of high added-value components from food wastes: Conventional, emerging technologies and commercialized applications. *Trends in Food Science & Technology*, 26(2), 68-87.

Galanakis, C. M. (2015). Chapter 3 - The universal recovery strategy *Food Waste Recovery* (pp. 59-81). San Diego: Academic Press.

Garde-Cerdán, T., Arias-Gil, M., Marsellés-Fontanet, A. R., Ancín-Azpilicueta, C., & Martín-Belloso, O. (2007). Effects of thermal and non-thermal processing treatments on fatty acids and free amino acids of grape juice. *Food Control*, 18(5), 473-479.

Gaub, V., & Hartgerink, J. D. (2008). Synthetic collagen heterotrimers: Structural mimics of wild-type and mutant collagen type I. *J Am Chem Soc*, 130(23), 7509-7515.

Glanbia.com. (2017). *Markets & Locations*. [online] Available at: <https://www.glanbia.com/our-business/global-performance-nutrition/markets-and-locations> [Accessed 5 Apr. 2017].

Global Market Insights report (2015). Collagen Market Size, Industry Analysis Report, Regional Outlook (U.S., Germany, UK, Italy, Russia, China, India, Japan, South Korea, Brazil, Mexico, Saudi Arabia, UAE, South Africa), Application Development Potential, Price Trend, Competitive Market Share & Forecast, 2016 – 2023. Available on <https://www.gminsights.com/industry-analysis/collagen-market> Accessed on February 2017.

Gómez-Guillén, M., Giménez, B., López-Caballero, M. a., & Montero, M. (2011). Functional and bioactive properties of collagen and gelatin from alternative sources: A review. *Food Hydrocolloids*, 25(8), 1813-1827.

Heinz, G., & Hautzinger, P. (2007). *Meat processing technology for small to medium scale producers*: FAO.

- Henchion, M, McCarthy, M & J. O'Callaghan (2016), Transforming beef by-products into valuable ingredients: which spell/recipe to use? *Front. Nutr. - Nutrition and Food Science Technology*, In press
- Hettiarachchy, N. S., Sato, K., Marshall, M. R., & Kannan, A. (2012). *Food proteins and peptides: chemistry, functionality, interactions, and commercialization*: CRC Press.
- Hickey, G. (2014) Bord Bia Food Alert: Sports Nutrition now more convenient than ever in Ireland. Available on: <http://www.bordbia.ie/industry/manufacturers/insight/alerts/Pages/SportsNutritionnowmoreconvenientthaneverinIreland.aspx?year=2014&wk=24>. Accessed on February 2017.
- Hoffmann, M., Waszkiewicz-Robak, B., & Świdorski, F. (2010). Functional food of animal origin. Meat and meat products. *Nauka Przyroda Technologie*, 4(5), # 63.
- Hsieh, Y.-H. P., & Ofori, J. A. (2011). Blood-derived products for human consumption. *Revelation and Science*, 1(01).
- Hurtado, S., Dagà, I., Espigulé, E., Parés, D., Saguer, E., Toldrà, M., & Carretero, C. (2011). Use of porcine blood plasma in "phosphate-free frankfurters". *Procedia Food Science*, 1, 477-482.
- Jayathilakan, K., Sultana, K., Radhakrishna, K., & Bawa, A. S. (2012). Utilization of byproducts and waste materials from meat, poultry and fish processing industries: a review. *J Food Sci Technol*, 49(3), 278-293.
- Kadam, S. U., Tiwari, B. K., Álvarez, C., & O'Donnell, C. P. (2015). Ultrasound applications for the extraction, identification and delivery of food proteins and bioactive peptides. *Trends in Food Science & Technology*, 46(1), 60-67.
- Kielty, C., & Grant, M. (2002). The collagen family: Structure, assembly, and organization in the extracellular matrix. In P. Royce & B. Steinmann (Eds.), *Connective Tissue and Its Heritable Disorders: Molecular, Genetic, and Medical Aspects* (Second ed., pp. 159-221). Hoboken, NJ, USA: John Wiley & Sons, Inc
- Korhonen, H., Pihlanto-Leppäla, A., Rantamäki, P., & Tupasela, T. (1998). Impact of processing on bioactive proteins and peptides. *Trends in Food Science & Technology*, 9(8), 307-319.
- Korhonen, H., & Pihlanto, A. (2006). Bioactive peptides: Production and functionality. *International Dairy Journal*, 16(9), 945-960..
- Kotch, F. W., & Raines, R. T. (2006). Self-assembly of synthetic collagen triple helices. *Proc Natl Acad Sci U S A*, 103(9), 3028-3033.
- Kumar, P., Satyam, A., Cigognini, D., Pandit, A., & Zeugolis, D. (In Press). Low oxygen tension and macromolecular crowding accelerate extracellular matrix deposition in human corneal fibroblast culture. *J Tissue Eng Regen Med*.
- Lee, S.-J., Kim, E.-K., Hwang, J.-W., Oh, H.-J., Cheong, S.-H., Moon, S.-H., . . . Park, P.-J. (2010). Purification and characterisation of an antioxidative peptide from enzymatic hydrolysates of duck processing by-products. *Food Chemistry*, 123(2), 216-220.
- Lemes, A. C., Sala, L., Ores, J. d. C., Braga, A. R. C., Egea, M. B., & Fernandes, K. F. (2016). A review of the latest advances in encrypted bioactive peptides from protein-rich waste. *International Journal of Molecular Sciences*, 17(6), 950.
- Leoci, R. (2014). *Animal by-products (ABPs): origins, uses, and European regulations*: Universitas Studiorum.
- Li, H., Zhu, K., Zhou, H., & Peng, W. (2012). Effects of high hydrostatic pressure treatment on allergenicity and structural properties of soybean protein isolate for infant formula. *Food Chemistry*, 132(2), 808-814.
- Linder, M., Fanni, J., Parmentier, M., Sergeant, M., & Phan-Tan-Luu, R. (1995). Protein recovery from veal bones by enzymatic hydrolysis. *Journal of Food Science*, 60(5), 949-952.
- Looy, H., Dunkel, F. V. & Wood, J. R. (2014). How then shall we eat? Insect-eating attitudes and sustainable foodways, *Agriculture and Human Values*, 31 (1), 131-141.
- López, M. A., & Martos, F. C. (2004). Iron availability: An updated review. *International Journal of Food Sciences and Nutrition*, 55(8), 597-606.

- Lynch, S., Mullen, A. M., O'Neill, E., & Álvarez, C. (2017). Harnessing the potential of blood proteins as functional ingredients. A review of the state of art in blood processing. *Comprehensive Reviews in Food Science and Food Safety*.
- Lynn, A. K., Yannas, I. V., & Bonfield, W. (2004). Antigenicity and immunogenicity of collagen. *J Biomed Mater Res B*, 71B(2), 343-354.
- Hickey, G. (2014) Bord Bia Food Alert: Sports Nutrition now more convenient than ever in Ireland. Available on: <http://www.bordbia.ie/industry/manufacturers/insight/alerts/Pages/SportsNutritionnowmoreconvenientthaneverinIreland.aspx?year=2014&wk=24>. Accessed on February 2017.
- King, H. (2011) Bord Bia Market report: Sports Nutrition - The European Market Publication Date: 23/06/2011. Available at: <http://www.bordbia.ie/industry/manufacturers/insight/publications/bbreports/Documents/Sports%20Nutrition%20-20The%20European%20Market.pdf>. Accessed on February 2017
- Marcet, I., Alvarez, C., Paredes, B., & Diaz, M. (2016). The use of sub-critical water hydrolysis for the recovery of peptides and free amino acids from food processing wastes. Review of sources and main parameters. *Waste Manag*, 49, 364-371.
- Marti, D. L., Johnson, R. J., & Mathews Jr, K. H. (2012). Where's the (not) meat? Byproducts from beef and pork production. *Journal of Current Issues in Globalization*, 5(4), 397.
- Martins, Y., Pelchat M.L. & Pliner P. (1997). Try it; it's good and it's good for you": effects of taste and nutrition information on willingness to try novel foods. *Appetite*. 28(2):89-102.
- Millward, D. J., Layman, D. K., Tomé, D., & Schaafsma, G. (2008). Protein quality assessment: impact of expanding understanding of protein and amino acid needs for optimal health. *The American journal of clinical nutrition*, 87(5), 1576S-1581S.
- Montero Castillo, P. M., Ligardo, M., Alejandro, Y., González, J., & Cristina, L. (2015). Protein Quality of Rice Drinks Fortified with Bovine and Porcine Blood Plasma. *Revista Facultad Nacional de Agronomía, Medellín*, 68(1), 7487-7496.
- Mullen, A.M., & Álvarez, C. (2016). Offal: types and composition. *The Encyclopedia of Food and Health*, 4, 152-157.
- Mullen, A. M., Álvarez, C., Pojić, M., Hadnadev, T. D., & Papageorgiou, M. (2015). Classification and target compounds. 25-57.
- Murray, S. M., Patil, A. R., Fahey, G. C., Merchen, N. R., & Hughes, D. M. (1997). Raw and rendered animal by-products as ingredients in dog diets. *Journal of animal science*, 75(9), 2497-2505.
- Myllyharju, J., Lamberg, A., Notbohm, H., Fietzek, P. P., Pihlajaniemi, T., & Kivirikko, K. I. (1997). Expression of wild-type and modified pro alpha chains of human type I procollagen in insect cells leads to the formation of stable [alpha 1(I)](2)alpha 2(I) collagen heterotrimers and [alpha 1(I)](3) homotrimers but not [alpha 2(I)](3) homotrimers. *J Biol Chem*, 272(35), 21824-21830.
- O'Leary, L. E., Fallas, J. A., Bakota, E. L., Kang, M. K., & Hartgerink, J. D. (2011). Multi-hierarchical self-assembly of a collagen mimetic peptide from triple helix to nanofibre and hydrogel. *Nat Chem*, 3(10), 821-828.
- O'Leary, L. E., Fallas, J. A., & Hartgerink, J. D. (2011). Positive and negative design leads to compositional control in AAB collagen heterotrimers. *J Am Chem Soc*, 133(14), 5432-5443.
- O'Sullivan, S., Lafarga, T., Hayes, M., & O'Brien, N. (2016). Potential bioactivity of hydrolysates derived from bovine lung. *Proceedings of the Nutrition Society*, 75(OCE3).
- Ofori, J. A., & Hsieh, Y.-H. P. (2012). *The use of blood and derived products as food additives*: INTECH Open Access Publisher.
- Olsen, D. R., Leigh, S. D., Chang, R., McMullin, H., Ong, W., Tai, E., . . . Toman, P. D. (2001). Production of human type I collagen in yeast reveals unexpected new insights into the molecular assembly of collagen trimers. *J Biol Chem*, 276(26), 24038-24043.

- Pares, D., Toldra, M., Saguer, E., & Carretero, C. (2014). Scale-up of the process to obtain functional ingredients based in plasma protein concentrates from porcine blood. *Meat Sci*, 96(1), 304-310.
- Pimentel, D., & Pimentel, M. (2003). Sustainability of meat-based and plant-based diets and the environment. *The American journal of clinical nutrition*, 78(3), 660S-663S.
- Power, O., Fernández, A., Norris, R., Riera, F. A., & FitzGerald, R. J. (2014). Selective enrichment of bioactive properties during ultrafiltration of a tryptic digest of β -lactoglobulin. *Journal of Functional Foods*, 9, 38-47.
- Rele, S., Song, Y., Apkarian, R. P., Qu, Z., Conticello, V. P., & Chaikof, E. L. (2007). D-periodic collagen-mimetic microfibers. *J Am Chem Soc*, 129(47), 14780-14787.
- Rivera, J. A. (1998) Petfood: physico-chemical characteristics and functional properties of meat by-products and mechanically separated chicken (MSC) in a high-moisture model system. Iowa State University, PhD Thesis. Retrospective Theses and Dissertations. Paper 11886.
- Rutschmann, C., Baumann, S., Cabalzar, J., Luther, K. B., & Hennet, T. (2014). Recombinant expression of hydroxylated human collagen in *Escherichia coli*. *Appl Microbiol Biotechnol*, 98(10), 4445-4455.
- Rozin, P. & Fallon, A. E. (1987), A perspective on disgust, *Psychological Review*, 94, 23-41
- Ryan, J. T., Ross, R. P., Bolton, D., Fitzgerald, G. F., & Stanton, C. (2011). Bioactive peptides from muscle sources: meat and fish. *Nutrients*, 3(9), 765-791.
- Sarmadi, B. H., & Ismail, A. (2010). Antioxidative peptides from food proteins: a review. *Peptides*, 31(10), 1949-1956.
- Satyam, A., Kumar, P., Cigognini, D., Pandit, A., & Zeugolis, D. (2016). Low, but not too low, oxygen tension and macromolecular crowding accelerate extracellular matrix deposition in human dermal fibroblast culture. *Acta Biomater*, 44, 221-231.
- Satyam, A., Kumar, P., Fan, X., Gorelov, A., Rochev, Y., Joshi, L., . . . Zeugolis, D. (2014). Macromolecular crowding meets tissue engineering by self-assembly: A paradigm shift in regenerative medicine. *Adv Mater*, 26(19), 3024-3034.
- Selmane, D., Christophe, V., & Gholamreza, D. (2008). Extraction of proteins from slaughterhouse by-products: Influence of operating conditions on functional properties. *Meat Sci*, 79(4), 640-647.
- Sharma, S., Singh, R., & Rana, S. (2011). Bioactive peptides: a review. *Int J Bioautomation*, 15(4), 223-250.
- Schmidt, C. (2014). The rise of protein in the global health and wellness and supplement arenas – Examining the global protein surge. Presented at the Protein Trends & Technologies Seminar, April 8-9, 2014, Arlington Heights, IL, USA. Available at <https://www.globalfoodforums.com/wp-content/uploads/2014/04/Chris-Schmidt-Euromonitor-2014-Protein-Trends-Technologies.pdf>. Accessed on February 2017.
- Smeller, L. (2002). Pressure–temperature phase diagrams of biomolecules. *Biochimica et Biophysica Acta (BBA)-Protein Structure and Molecular Enzymology*, 1595(1), 11-29.
- Soliva-Fortuny, R., Balasa, A., Knorr, D., & Martín-Belloso, O. (2009). Effects of pulsed electric fields on bioactive compounds in foods: a review. *Trends in Food Science & Technology*, 20(11), 544-556.
- Sorushanova, A., Delgado, L., Wu, Z., Shologu, N., Kshirsagar, A., Raghunath, R., . . . Zeugolis, D. (Submitted). New tricks for the old protein - The next generation of collagen-based devices. *Prog Polym Sci*.
- Steen, L., Glorieux, S., Goemaere, O., Brijs, K., Paelinck, H., Foubert, I., & Fraeye, I. (2016). Functional properties of pork liver protein fractions. *Food and Bioprocess Technology*, 9(6), 970-980.
- Stein, H., Wilensky, M., Tsafrir, Y., Rosenthal, M., Amir, R., Avraham, T., . . . Shoseyov, O. (2009). Production of bioactive, post-translationally modified, heterotrimeric, human recombinant type-I collagen in transgenic tobacco. *Biomacromolecules*, 10(9), 2640-2645.
- Sun, L. (2013). Peptide-based drug development. *Modern Chemistry & Applications*, 2013.

- Thomas, D., Gaspar, D., Soroushanova, A., Milcovich, G., Spanoudes, K., Mullen, A.M., . . . Zeugolis, D. (2016). Scaffold and scaffold-free self-assembled systems in regenerative medicine. *Biotechnol Bioeng*, 113(6), 1155-1163.
- Toldra, F., Aristoy, M. C., Mora, L., & Reig, M. (2012). Innovations in value-addition of edible meat by-products. *Meat Sci*, 92(3), 290-296.
- Toldrá, F., Mora, L., & Reig, M. (2016). New insights into meat by-product utilization. *Meat science*, 120, 54-59.
- Toman, P. D., Pieper, F., Sakai, N., Karatzas, C., Platenburg, E., de Wit, I., . . . Platenburg, G. J. (1999). Production of recombinant human type I procollagen homotrimer in the mammary gland of transgenic mice. *Transgenic Res*, 8(6), 415-427.
- Tomita, M., Munetsuna, H., Sato, T., Adachi, T., Hino, R., Hayashi, M., . . . Yoshizato, K. (2003). Transgenic silkworms produce recombinant human type III procollagen in cocoons. *Nat Biotechnol*, 21(1), 52-56
- Trostle, R. (2008). *Global agricultural supply and demand: factors contributing to the recent increase in food commodity prices*: US Department of Agriculture, Economic Research Service Washington, DC.
- USDA-FSIS Directive 7120.1. (2017) Safe and suitable ingredients used in the production of meat, poultry, and egg products. Revision 39, 1/9/17
- USDA-FSIS (1999). Saving of blood from livestock as an edible product. *Federal Register* / Vol. 64, No. 246 Part 9 CFR 310.20
- Van, D. M. P. C. (2001). Food products processed from offal components: Google Patents.
- Van Huis, A., Van Itterbeeck, J., Klunder, H., Mertens, E., Halloran, A., Muir, G., & Vantomme, P. (2013). *Edible insects: future prospects for food and feed security*: Food and agriculture organization of the United nations (FAO).
- Venegas Fornias, O. (1996). Edible by-products of slaughter animals. *FAO Animal Production and Health Paper (FAO)*.
- Vernooij, A., (2012) The Return of Animal By-Products. Rabobank Industry Note #335 –Dec 2012. Downloaded from <http://ausrenderers.com.au/index.php/downloads/category/1-general-documents> 7th Feb 2017
- Vuorela, A., Myllyharju, J., Nissi, R., Pihlajaniemi, T., & Kivirikko, K. I. (1997). Assembly of human prolyl 4-hydroxylase and type III collagen in the yeast *pichia pastoris*: Formation of a stable enzyme tetramer requires coexpression with collagen and assembly of a stable collagen requires coexpression with prolyl 4-hydroxylase. *EMBO J*, 16(22), 6702-3712.
- Waldron, K. (2007). *Waste minimization, management and co-product recovery in food processing: an introduction* (Vol. 1): Woodhead Publishing Limited: Cambridge, UK.
- Williams, P. (2007). Nutritional composition of red meat. *Nutrition & Dietetics*, 64(s4), S113-S119.
- Wilson, Marc (2011). The probable value of the US dried animal blood plasma market is \$29 million per annum. Global Advisors, online article published on 5 December 2011. Available at: <http://www.globaladvisors.biz/fast-facts/20111205/the-probable-value-of-the-us-dried-animal-blood-plasma-market-is-29-million-per-annum/>. Accessed on February 2017
- Zeugolis, D. I., Paul, R. G., & Attenburrow, G. (2008). Factors influencing the properties of reconstituted collagen fibers prior to self-assembly: animal species and collagen extraction method. *J Biomed Mater Res A*, 86(4), 892-904.
- Zeugolis, D. I., Paul, G. R., & Attenburrow, G. (2009). Cross-linking of extruded collagen fibers—A biomimetic three-dimensional scaffold for tissue engineering applications. *Journal of Biomedical Materials Research Part A*, 89(4), 895-908.
- Zhang, C., Baez, J., Pappu, K. M., & Glatz, C. E. (2009). Purification and characterization of a transgenic corn grain-derived recombinant collagen type I alpha 1. *Biotechnol Prog*, 25(6), 1660-1668.

847 Zou, Y., Wang, L., Li, P., Cai, P., Zhang, M., Sun, Z., . . . Xu, X. (2017). Effects of ultrasound assisted
848 extraction on the physiochemical, structural and functional characteristics of duck liver
849 protein isolate. *Process Biochemistry*, 52, 174-182.
850

851

852

853

854

855

856

857

858

859

860

861

862

863

864

865

866

Tables

Table 1: Total protein content (% wet basis), individual essential amino acids (g/100 g protein) and total essential amino acids (%EAA) in selected offal across a number of species.

Product	Specie	Prot. (%)	Lys	Met	Trp	% EAA
Blood	Cattle	17-18	9.7	2.4	-	60.6
	Porcine	18.5	9.0	2.3	1.5	58
Brain	Cattle	10.5	6.0	2.1	4.7	42.8
	Porcine	10.3	7.8	2.0	4.2	46.8
	Sheep	12.3	6.4	2.0	1.1	42.8
Heart	Cattle	17	8.2	2.6	1.1	47.1
	Porcine	17	8.3	2.6	1.2	47.7
	Sheep	18	7.5	2.2	1.1	43.8
Kidney	Cattle	15.3	6.6	2.1	1.4	45.2
	Porcine	15.4	7.2	2.1	1.3	48.0
	Sheep	18.0	6.5	2.0	1.4	43.9
Liver	Cattle	21	6.9	2.5	1.4	49.1
	Porcine	19	7.7	2.5	1.4	48.9
	Sheep	20.3	5.4	2.1	1.2	42.7
Lung	Cattle	17	7.1	2.0	0.9	41.5
	Porcine	15	7.3	1.6	0.9	37.8
	Sheep	12.5	6.5	1.8	0.9	40.6
Spleen	Cattle	19	7.2	1.8	1.0	48.0
	Porcine	17.9	7.5	1.8	1.0	41.9
	Sheep	17.2	7.7	1.9	1.1	48.5
Tongue	Cattle	17.1	7.7	2.1	0.8	45.8
	Porcine	16.3	8.2	2.2	1.2	40.2
	Sheep	15.3	7.1	2.1	1.0	40.4
Connective tissue collagen		85%	3.5	0.7	0.0	11.97

Data from Venegas, 1996.

878 Table 2: Summary of bioactive compounds from meat sources

Compound	Concentration (mg/g)	Biological function
Conjugated linoleic acid	3-8 (beef fat)	Reduced risk of colorectal cancer and diabetes, antioxidant, immunomodulatory.
Carnosine	2.7 (pork shoulder)	Antioxidant
Anserine	nd	Antioxidant, metal chelating
L-carnitine	1.3 (beef tight)	Lowning-cholesterol, calcium absorption, prevention of muscle myopathy
Glutathione	0.12-0.26 (beef muscle)	Antioxidant, defence against toxicological and pathological processes
Taurine	0.77 (beef muscle)	Eye health, preventing heart disease, essential in some stages(lactation or immune challenge)
Coenzyme Q10	0.02 (beef muscle)	Antioxidant
Creatine	3.5 (beef muscle)	Enhancing muscle performance

*Adapted from (Arihara and Ohata (2008))

894 Table 3: Main Regulations and Directives regarding the use of meat processing co-products
895 in Europe

Scope	Reference	Short description
General	Reg.(EC) 999/2001	rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies in animals
	Directive 2008/98/EC	concepts and definitions related to waste management, such as definitions of waste, recycling, recovery
	Directive 2008/1/EC	integrated pollution prevention and control
	Reg.(EC) 1069/2009	health rules as regards animal by-products and derived products not intended for human consumption
For human consumption	“Hygiene Package”	
	· Reg.(EC) 852/2004	on the hygiene of foodstuffs;
	· Reg.(EC) 853/2004	specific hygiene rules for food of animal origin;
	· Reg.(EC) 854/2004	specific rules for products of animal origin intended for human consumption
	Reg.(EC) 1169/2011	provision of food information to consumers
	Reg.(EC) 609/2013	food intended for infants and young children, food for special medical purposes, and total diet replacement for weight control
	Reg.(EC) 1333/2008	approved food additives and their conditions of use
Specific applications	Reg.(EC) 1332/2008	enzymes used in food
	Directive 76/768/EEC	cosmetic products
	Directive 90/385/EEC	medical implants
	Directive 2003/32/EC	medical devices
	Directive 89/79/EC	<i>In vitro</i> diagnostic medical devices
	Directive 2001/82/EC	vet medical products
	Directive 2001/83/EC	medicinal products

896

897

898

899

900

901

902

903

904

905

Product	Revalorizing techniques	High added-value products
Liver	Enzymatic hydrolysis	Antioxidant peptides
Heart	Isoelectric solubilization/precipitation	High value protein with low ash, fat and cholesterol
	Phosphate buffer washing	Myofibrillar concentrate as texturizing
Skin	Collagen recovery	Barrier membrane, drug delivery, fibroblast scaffolds, bioengineered tissues
	Enzymatic hydrolysis and chromatographic purification	Antioxidant peptides and liver protectors
	Collagen hydrolysis	Antioxidant activity, antimicrobial properties antihypertensive, biomimetic tissue
Blood	Enzymatic hydrolysis	Antioxidant, antibacterial, antihypertensive or iron-binding peptides
	Chemical hydrolysis	Pre-digested peptides for animal and pet food
	Ethanol precipitation	Purified protein as food ingredient
	High pressure treatment	Peptides and biopreserved blood
		Amino acid and peptides production
Bone	Subcritical water hydrolysis	Hydroxyapatite and collagen
	Subcritical water	New kind of sausages
	Alkaline extraction	Protein concentrates with good functional properties
Lung	Isoelectric solubilization - precipitation and membrane filtration	
Feathers and hair	Keratinolytic bacteria fermentation and enzymatic hydrolysis	Keratinolytic protease production, culture medium, soil assessment, separation membranes